

CellOptimizer Final Report

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1.0 Executive Summary

This document seeks to describe the background information, customer requirements, design specifications, indications for use, selected materials, proposed budget, prototypes, final design, manufacturing processes, and testing methods regarding the *CellOptimizer* automated microscope stage product.

2.0 Introduction and Background

There is currently a need to optimize the imaging process for 16-well microfluidic chips in Dr. Hawkins' Laboratory. In order to efficiently analyze the effect of varying culture conditions over time, an automated system must be developed to capture clear images of each well on the chip at specified time points. The hardware component of this system will consist of a microscope stage with mounted actuators for precise position control in the x and y direction. The software component of the system will incorporate an Arduino code to synchronize the automatic image capture of the LabSmith SVM-340 inverted microscope with the position of the stage. This synchronization will enable users to capture images of all 16 wells over a several-day period of time with no user-input required. Ultimately, this project will yield a device that will enable the optimization of cell culture data collection for long-term NIH 3T3 co-culture systems.

3.0 Customer Requirements and Design Specifications

3.1 IFU

The *CellOptimizer* automated microscope stage is intended for use with the LabSmith SVM-340 Microscope and 16-well NIH 3T3 microfluidic chips in Dr. Hawkins' Laboratory. For a user-defined period of time, this device will automatically position each of the 16 wells on the provided microfluidics chip in the field of view of the LabSmith SVM-340, allowing the microscope to capture a specified number of images of each well with no additional user input.

3.2 Product Design Specifications

The Product Design Specifications, including the customer requirements and associated design specifications, are summarized below in Table I. The first requirement describes that the CellOptimizer must have automated, motorized position control in the xy plane. The corresponding design specification details that the stage must be able to move at least 25 mm and 15 mm in the x and y direction, respectively. These dimensions correspond to the length and width of the microfluidics chip that the CellOptimizer will need to position over the LabSmith SVM-340 for comprehensive culture well imaging. The second requirement specifies that the machined microscope stage must be compatible with the LabSmith SVM-340. There is a circular peg on each corner of the rectangular LabSmith Microscope. The CellOptimizer must be less than 21 cm x 27 cm in order to fit over these pegs and remain stable during actuator movement. The third customer requirement is that the actuators responsible for displacing the microscope stage in the x and y direction must have high position accuracy. Specifically, in order to ensure that the stage positions each culture well in the field of view of the LabSmith Microscope, the actuators must have position accuracy within 5 microns. Furthermore, the fourth customer requirement specifies that stage position error must not be a function of actuator displacement. In other words, the position of the accuracy of the stage must not significantly fluctuate with distance travelled. This is critical to the success of the device, because image accuracy must be consistent across each column and row of culture wells on the microfluidics chip. If accuracy decreases with displacement, comparisons between culture wells at either end of the microfluidics chip will likely be inconclusive or misleading. Lastly, the stage must have a position return repeatability within 1 micron. This is necessary because the microscope must be able to take multiple images of each culture well over a specified period of time. If the stage positions a different area of the culture well in the field of view of the microscope each time it is imaged, it will be difficult to make accurate comparisons between time points for each well.

Table I. CellOptimizer Product Design Specifications

Customer Req.	Engineering metric	Specification	Rationale
Motorized position and computer control in x and y direction	Displacement (d)	x-direction: $d > 25 \mu\text{m}$ y-direction: $d > 15 \mu\text{m}$	The stage must be able to move across the entire length and width of the microfluidic chip in order to capture an image of each culture well
Stage must be compatible with LabSmith SVM-340 Microscope	Geometry	Stage must be less than 21 cm wide and less than 27 cm long	LabSmith SVM-340 is 21 cm x 27 cm
High position accuracy	Accuracy	Position accuracy within $5 \mu\text{m}$	High position accuracy required to ensure that the culture wells consistently fall within the field of view of the microscope during image capture.
Position error is not a function of actuator displacement	Accuracy	Position accuracy does not significantly vary with displacement of the actuator	If position accuracy decreases with displacement or vice versa, culture well images at one end of the chip will be more accurate than those captured at the other end. This will make comparisons between different culture wells challenging and potentially misleading.
High position return repeatability	Repeatability	Position return repeatability within $1 \mu\text{m}$	The microscope must be able to take multiple images of the same culture well over a specified period of time. To ensure accurate comparison of different images of the same well over time, it is important that the same portion of the well is in the field of view.

3.3 House of Quality

Quality Function Deployment (QFD) methodology was implemented to ensure that the customer requirements outlined in Section 3.0 were reflected in the design constraints of the CellOptimizer device.

3.3.1 HOQ Room 1

As illustrated below in Table II, all customer requirements were assigned an importance ranking of 5, with the exception of cost. This is because each customer requirement was explicitly tied to the success of final design. For example, if the stage does not have motorized position control in the x and y direction, the device will be unable to properly position each culture well over the LabSmith SVM-340 field of view for image capture. Furthermore, if the stage position control is not accurate and repeatable, the LabSmith SVM-340 will be unable to capture clear, comparable images of the culture wells over time. Cost was assigned a lower importance ranking because this project had a relatively flexible budget.





Table II. Room 1 - Customer Requirements and the associated importance rankings

Customer Requirements	Importance Ranking
Motorized position and computer control in x and y direction	5
Compatible with LabSmith SVM-340 Microscope	5
Position error does not significantly fluctuate with actuator displacement	5
High position accuracy	5
High position return repeatability	5
Cost	2

3.3.2 HOQ Room 2

The engineering metrics associated with each customer requirement are outlined below in Table III. An increase in displacement in the xy plane, position accuracy, and position return repeatability, as well as a decrease in stage geometry was desired. An increased displacement in the xy plane was preferred because the stage must be able to move at least 25 mm and 15 mm in the x and y direction, respectively. A greater displacement will not impede the function of the final design, while a smaller displacement will prevent the LabSmith SVM-340 from capturing images of every culture well on the microfluidics chip. Additionally, a greater position accuracy and position return repeatability will yield culture well images that are comparable across different time points and culture conditions. This will ultimately allow Dr. Hawkins' laboratory to confidently conclude which culture conditions are optimal for a given circumstance. It is important to note that the improvement direction for position error as a function of displacement was intentionally left blank, as position error should ideally remain constant over the entire range of actuator displacement.

Table III. Room 2 - Engineering Characteristics and desired direction of improvement

Engineering Characteristics				
		-		
mm	cm	mm	μm	μm
Displacement in xy plane	Stage Geometry	Position Error(Displacement)	Position Accuracy	Position return repeatability

3.3.3 HOQ Room 3

The matrix presented in Table IV below describes the strength of the relationship between each customer requirement and engineering metric. A value of 0, 1, 3, or 9 represents a non-existent, weak, moderate, or strong relationship, respectively.

Table IV. Room 3- Relationship matrix between customer requirements and engineering characteristics

	Engineering Characteristics				
Customer Requirement	Displacement in xy plane	Stage Geometry	Position Error(Displacement)	Position Accuracy	Position Return Repeatability
Motorized position control (xy)	9	0	9	9	9
Compatible with LabSmith SVM-340	0	9	0	0	0
Position Error does not significantly fluctuate with actuator displacement	3	0	9	9	3
High position accuracy	3	0	9	9	3
High Return repeatability	3	0	3	3	9
Cost	3	3	9	9	9

3.3.4 HOQ Room 5

As displayed in Table V below, position error over actuator displacement and position accuracy tied for the most important engineering characteristics, followed by position return repeatability, displacement in the xy plane, and stage geometry. Due to the high rank of position error over actuator displacement, accuracy, and repeatability, these engineering characteristics were used as decision-making criteria for evaluating candidate designs.

Table V. Room 5 - Relative importance ranking of each engineering characteristic

	Engineering Characteristics				
	Displacement in xy plane	Stage Geometry	Position Error(Displacement)	Position Accuracy	Position return repeatability
Raw Score	96	51	168	168	138
Relative Weight	0.16	0.082	0.27	0.27	0.22
Rank Order	3	4	1	1	2

3.3.5 HOQ Room 6

The customer assessment of competing products is summarized below in Table VI. The Zaber Motorized XY Microscope Stage, MLS203-1 Thorlabs, MS-2000 XYZ, and CellOptimizer were ranked on a scale of 1 to 5, with 5 being the highest, for each of the customer requirements. The CellOptimizer received the highest overall ranking, confirming that this device successfully meets each customer requirement.

Table VI. Room 6 - Customer Assessment of Competing Products

Customer Req.	Customer Assessment of Competing Qualities			
	Zaber Motorized XY Microscope Stage	MLS203-1 Thorlabs	MS-2000 XYZ	<i>CellOptimizer</i>
Motorized position control (xy)	5	5	5	5
Compatible with LabSmith SVM-340	1	1	1	5
Position Error does not significantly fluctuate with actuator displacement	3	3	5	5
High position accuracy	3	5	5	5
Return repeatability	5	5	3	5
Cost	1	1	1	3

4.0 Stage Gate Process

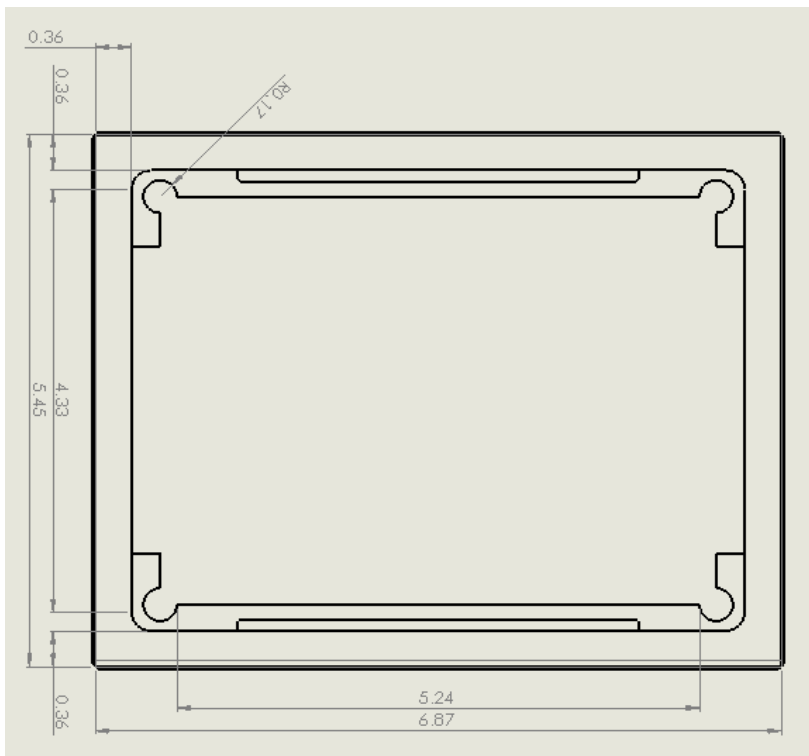
4.1 Concept Review

This automated stage is being designed as an in-house solution that is compatible with the SVM340 microscope. The stage will need to be able to position each of the 16 wells on a microfluidic chip over a camera. The translational movement requirements are more than 25 mm in the x direction and more than 15 mm in the y direction. The automated stage will need to recognize when each well is over the camera so that a picture of the cells can be captured. For the current scope of the project, the total market is limited to people who have SVM340 microscopes and who use the exact same microfluidic chip design as Dr. Hawkins. Because this

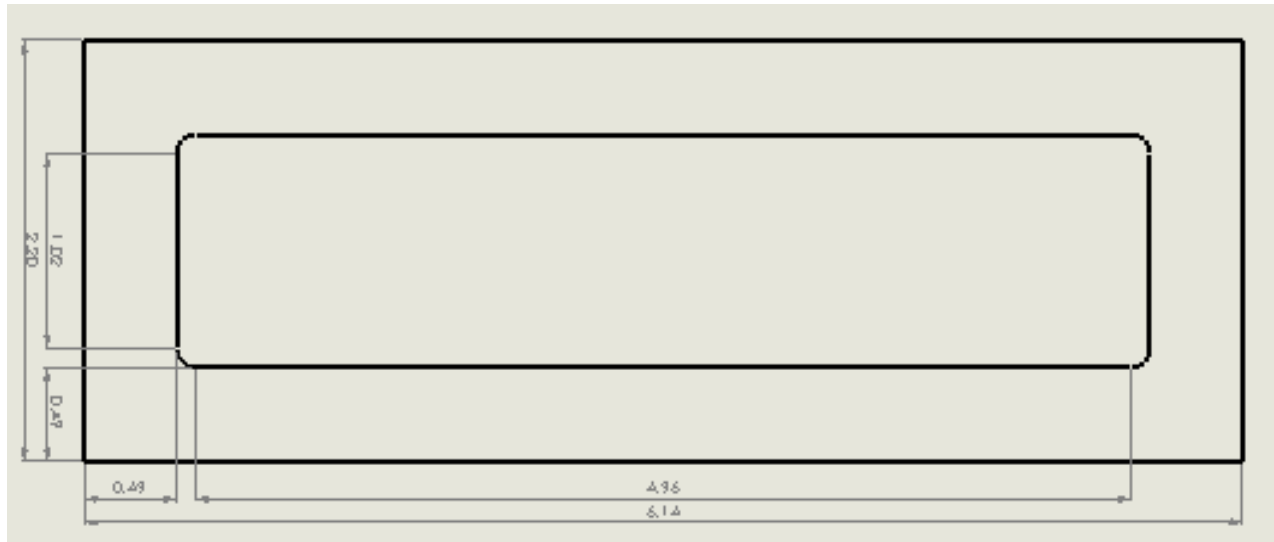
product is an in house solution, the specifications are more specific than what may be useful for a bigger market.

4.2 Design Freeze

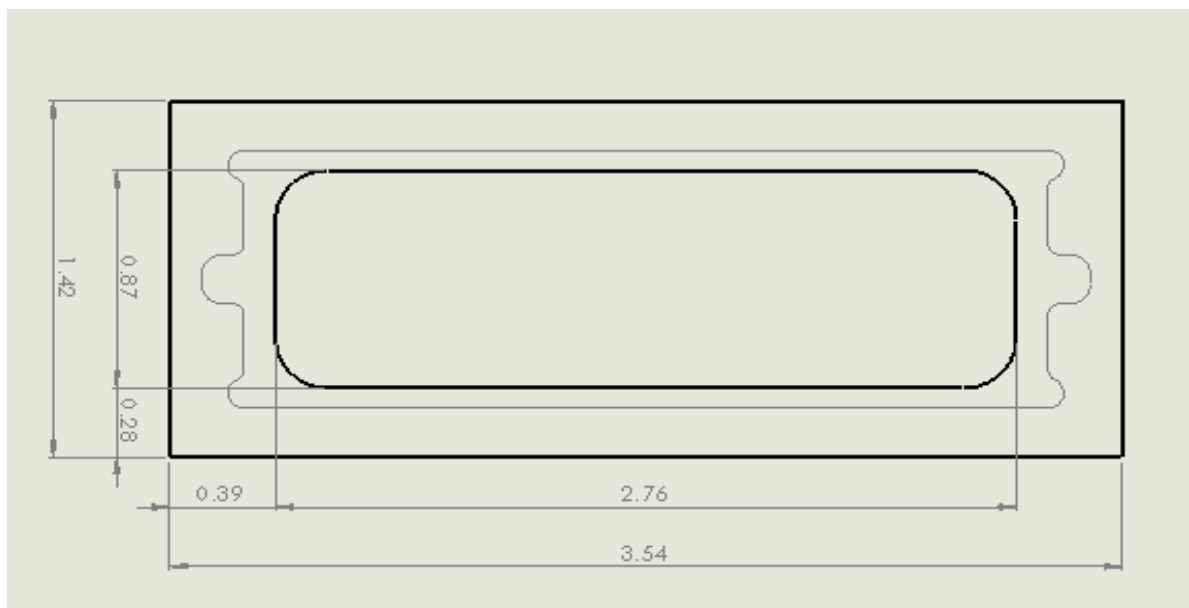
Outer Stage: The outer stage is made out of 6061 aluminum and has outer dimensions of 5.45"x6.87". The outer stage is meant to fit on a SVM340 inverted microscope via a peg system that is already implemented on the microscope. The outer stage will house the inner stage and allow the inner stage to traverse in the x-direction.



Inner Stage: The inner stage is made out of 6061 aluminum and has outer dimensions of 6.14"x2.2". The inner stage will house the slide holder and allow the slide holder to move in the y-direction.



Slide Holder: The slide holder is made out of 6061 aluminum and has outer dimensions of 3.54" by 1.42". The slide holder is meant to house a standard 2.95"x.98" microscope slide with a microfluidic chip adhered to the slide.



4.3 Design Review

The automated microscope stage is comprised of an outer stage, an inner stage, and a slide holder. All 3 parts are constructed from 6061 aluminum with a CNC and mill. The outer stage is the attachment point to the SVM340 microscope and supports the inner stage in translational movement in the x-direction. The inner stage supports the translational y-direction movement of the slide holder. The slide holder supports a standard 2.95"x.98" microscope slide with a custom microfluidic device on the slide.

5.0 Description of Final Prototype Design

5.1 Overview

The final prototype design includes 3 stages: an outer stage for x-axis movement, an inner stage for y-axis movement, and a slide holder for securing the microchip. Each stage component has a pushing block and mounting block to interface with the discontinued Newport 850G linear actuator. The outer and inner stages have lips that allow the interior parts to rest inside and travel along their required axis. The outer stage has four holes that align with pegs on the Labsmith SVM-340.

5.2 Design Justification

The design fits into place on the LabSmith SVM340 and the aluminum is heavy enough to support the weight of the actuators when both actuators are attached. Additionally, the actuators have enough strength to push the components they are attached to. The design allows enough travel in the x and y directions for all 16 wells on the microfluidic device to be imaged by the microscope. The design is portable and the actuators no longer need to be controlled by the large Newport Controller, as they are connected to an Arduino.

5.3 Analysis

The actuators were able to successfully be controlled by the Arduino. This is a huge improvement over the Newport Controller because the controller itself is too large to feasibly be used with the SVM340 microscope. Controlling the actuators with the Arduino allows for a portable stage system with various coded inputs for the actuators. After some testing, the force required to push the slide towards the actuator was achieved by using a rubber band around the mounting and pushing block. The rubber band applies a constant force pushing the slide into the actuator so when the actuator recedes, the slide follows.

5.4 Cost Breakdown

The cost of each hardware and software component of the CellOptimizer device is summarized below in Table VII.

Table VII. CellOptimizer B.O.M.

Item / Quantity	Product Number	Vendor	Purpose	Price
6061-T651 Aluminum (2)	ASTM B209	Midwest Steel and Aluminum	Raw stage material; 2" x 6" x 7"	\$60 / block
Newport 850G Series Linear Actuator (2)	N/A (Discontinued)	Newport Corporation	Provide motion	\$350 / each
Newport AB-4 Actuator Pushing Block (2)	AB-4	Newport Corporation	Interface with actuator	\$22 / each
Newport AB-3 Actuator Mounting Block (2)	AB-3	Newport Corporation	Mount actuators	\$21/ each
CNC Machining	N/A	N/A	Machine outer stage & slide holder	\$300
Arduino Uno (1)	ATMega328P	Arduino	Controls actuator	\$22 / each
L923D IC MTRDRV BIPLR (8)	497-2936-5-ND	DigiKey Electronics	Protects Arduino from Actuator Current	\$8.59 / 8 pieces
Kuman Arduino Kit	K4-US		Jumper wires to connect breadboard to	\$29.29 / each

			arduino and terminal blocks	
DB25 25-pin Female Adapter RS-232 Serial Port Interface Breakout Board Connector (6)	RS-232	Amazon	Connects actuators to breadboard	\$7.93 / 6 pieces
¾ in. Zinc Plated Corner Braces (3)	SKU #527580	Home Depot	Build up pushing block attached to slide holder	\$1.97 / package
#4-40 x ½ in. Phillips Round-Head Machine Screws (1)	SKU #749848	Home Depot	Attach Mounting Block to Outer-Stage	\$0.44 / each
#4-40 x ¼ in. Phillips Round-Head Machine Screws / (1)	SKU #963277	Home Depot	Attach pushing block to slide holder via door hinge	\$0.66 / 2-pack
#4-40 x ½ in. self-tapping screw (1)	MS-ST-4-40	TUBEDEPOT	Attach Pushing Block to Inner-Stage	\$0.16 / each
Gorilla Glue Epoxy	4200101	Gorilla	Attach pushing/ mounting blocks to stage	\$5.47 / bottle
Testing	N/A	N/A	Misc: Duct tape, calipers, tools	\$29.98
Total				\$1,312.49

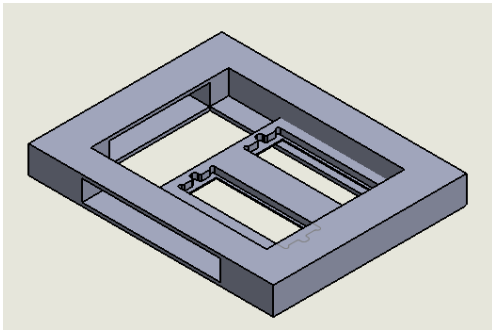
5.5 Safety Considerations

The automated stage microscope is a relatively safe device with few safety considerations. The reagents used within the microfluidic chip may contain toxic organic compounds and should be handled according to standard lab safety protocol. The voltage from the Arduino and power supply are small, but electronics should still be handled carefully. Finally, the stage itself is manufactured from aluminum and has caused injury after falling. Precautions should be taken to properly fix the stage to the microscope.

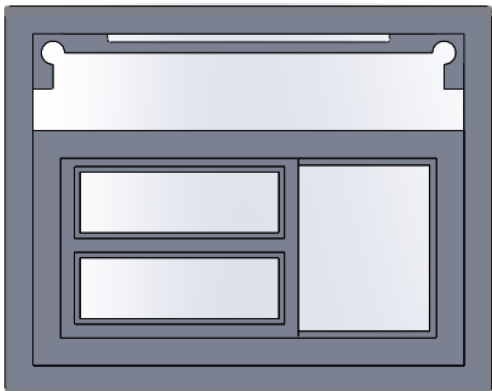
6.0 Prototype Development

6.1 Model Analyses

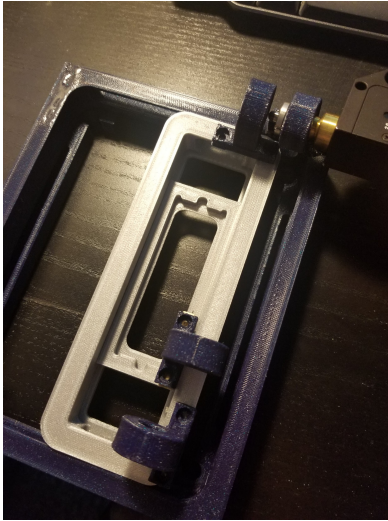
1. Prototype 1: This PLA 3D printed prototype was used to ensure proper fit between the Labsmith 340 and the outer stage and slide holder. Although the interface with the microscope had a proper fit, the slide holder was not able to fit inside the inner stage.



2. Prototype 2: This PLA 3D printed prototype was used to ensure proper fit between the Labsmith 340 and the outer stage, inner stage, and slide holder. All parts fit together with proper fit, though there was no lip or rail system to keep the inner stage inside the outer stage. The design did not meet the displacement specification, as the two-slide design did not allow enough room.



3. Prototype 3: This final 3D printed prototype had a proper fit between all three parts and were compatible with the Labsmith 340 microscope. This design had 3D printed pushing and mounting blocks to test compatibility with the full system. The designs were adjusted to allow more room for the microfluidic chip in the slide holder and smaller slot on the outer stage to prevent buckling. Additionally, the corners were filleted to ensure manufacturability.
- 4.



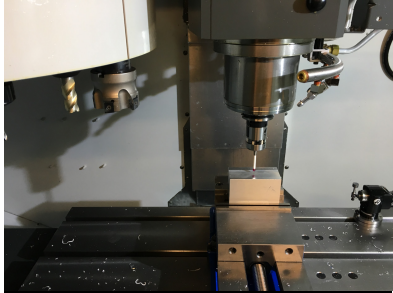

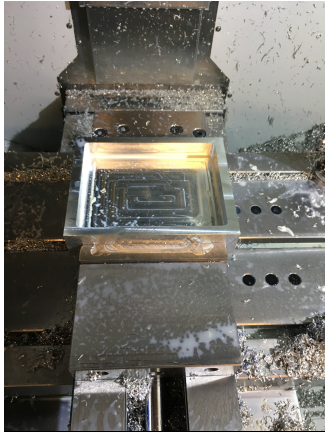
6.2 Evolution of Prototypes


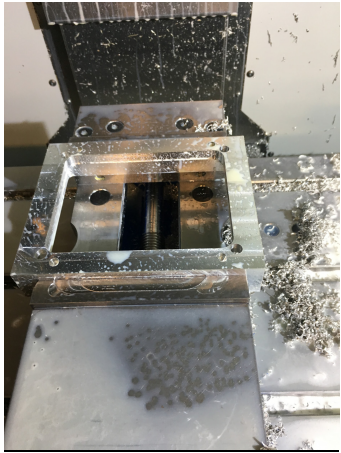
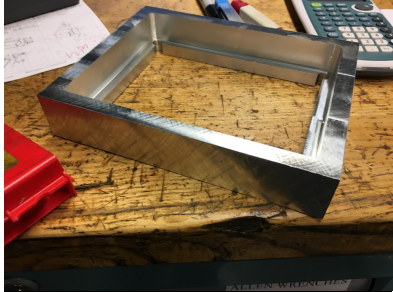
1. Prototype 1: Although the interface with the microscope had a proper fit, the slide holder was not able to fit inside the inner stage. The tolerances on the design were reevaluated to ensure proper fit.
2. Prototype 2: All parts fit together, with some necessary sanding due to 3D print-tolerancing. There was no lip or rail system to keep the inner stage from falling inside the outer stage. The design did not meet the displacement specification, as the two-slide design did not allow enough room.
3. Prototype 3: The designs were adjusted to allow more room for the microfluidic chip in the slide holder and smaller slot on the outer stage to prevent buckling. Additionally, the corners were filleted to ensure manufacturability. Rubber bands were added to ensure bi-directional movement for each actuator. Prototype 3 was used as a final design for the CNC and hand-milled manufacturing process.

6.3 Manufacturing Process




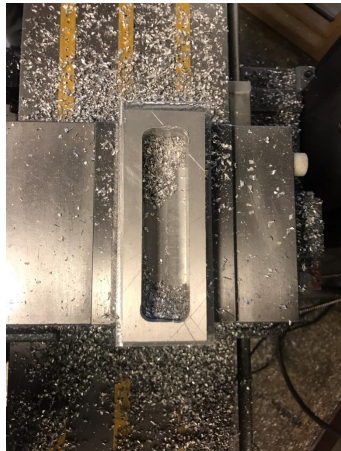
*Bill of Materials: Refer to *Table VIII*




Outer Stage Process

Step	Instructions	Picture
1	Start with a 7" x 6" x 2" block of 6061 Aluminum and put it in the CNC	
2	Cut the block down to the appropriate dimensions of 5.45" x 6.87"	
3	Cut out the middle of the block, leaving a .5" thick border around the stage	




4	Cut a hole that goes all the way through the part which leaves a .1" lip around the inside of the stage	
5	Flip the piece on its top side and drill the 4 holes in the bottom where the stage connects to the microscope	
6	Clean the outer stage with simple green and deburr the edges	



Inner Stage Process

Step	Instructions	Pictures
1	Start with a 7"x6"x2" Aluminum block and use a horizontal band saw to cut it to 7"x3"x2"	
2	Use a 5/8" endmill at a cutting speed of 660 on the mill to cut the block to 6.14" x 2.2"	
3	Use a 5/8" endmill at 660 cutting speed to cut a hole in the inner stage that is 1.26" x 4.96" and is .48" in from each of the sides	
4	Use a 5/8" endmill at a cutting speed of 660 to cut a hole through the inner stage that is .1" smaller on all sides than the cut made in Step 3. This will make the lip for the slide holder to rest on	

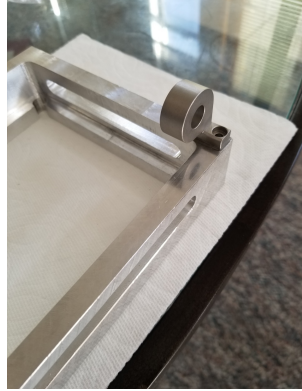

5	Use a vertical bandsaw to cut the inner stage from the bigger block of aluminum	 <p>A photograph showing a person's hands guiding a rectangular block of aluminum through a vertical bandsaw. The saw is mounted on a metal stand, and the block is being cut into two pieces. The background shows a workshop environment with various tools and equipment.</p>
6	Use a belt sander to smooth out the sharp edges and remove tooling marks	 <p>A photograph showing a person's hands using a belt sander to smooth a piece of aluminum. The sander is a Wilton brand machine with a red sanding belt. The person is holding the aluminum block against the belt, and the machine is mounted on a metal stand. The background shows a workshop environment.</p>
7	Use a fly cutter endmill at a cutting speed of 660 to remove .01" of material on all the faces and give the inner stage a smooth finish	 <p>A photograph showing a fly cutter endmill machining a piece of aluminum. The machine is a vertical lathe or mill, and the endmill is cutting the aluminum block. The background shows a workshop environment with various tools and equipment.</p>



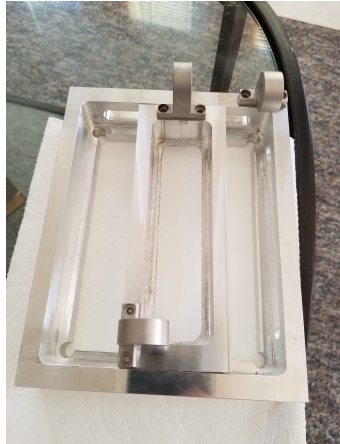
Slide Holder Process

Step	Instructions	Pictures
1	Use the CNC to machine a piece of aluminum down to 3.54"x1.42"x.25"	 A photograph showing a CNC machine in the process of cutting a rectangular piece of aluminum. The machine's tool is visible, and a fine mist of coolant is being applied to the cutting area. The workpiece is held in a fixture.
2	Cut a hole that is 2.76"x.87" and is .39" and .28" away from each side respectively	 A close-up photograph of the CNC machine cutting a hole into the aluminum piece. The machine's tool is positioned to create a rectangular slot. The workpiece is held in a fixture.
3	Cut a hole through the piece leaving a .1" lip from the cut made in Step 2	 A photograph showing the finished aluminum piece with a rectangular hole. The hole is slightly offset from the center, leaving a small lip on one side. The piece is held in a fixture.

4	Clean the part with simple green and deburr it	
5	Use a 3/16" endmill at a cutting speed of 660 on the mill to remove the fillet radius made by the CNC machine so that a 2.95"x.98" slide can fit	

Final Hardware Assembly

<u>Step</u>	<u>Instructions</u>	<u>Pictures</u>
1	Use gorilla glue epoxy to secure one of the mounting blocks to the corner of the outer stage	
2	Use a 1/4 in. endmill to drill hole in the center of the inner stage and outer stage. Tap each hole using a #4-40 HSS hand tap.	
3	Screw #4-40 1/4 in. Phillips Machine screw through the hole on the mounting block into the hole in the outer stage.	
4	Screw #4-40 1/4 in. Phillips Machine screw through the hole on the pushing block into the hole in the inner stage	
5	Screw #4-40 1/4 in. Phillips self tapping screw through the hole in second mounting block into the corner of the inner stage.	

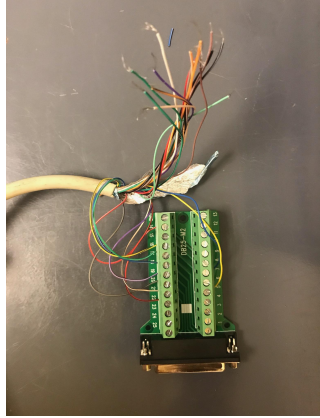

6	<p>Use the epoxy to glue two Zinc Plated Corner Braces together. Glue third corner Brace (with right angle facing upward) on the side of the other braces. Epoxy bottom corner braces to edge of slide holder. Secure with C-clamp and let set overnight.</p>	
8	<p>Use #4-40 1/2 in. Phillips Machine Screws to secure the second pushing block onto the top corner brace.</p>	
4	<p>Place the inner stage into the outer stage so that the mounting and pushing blocks align</p>	

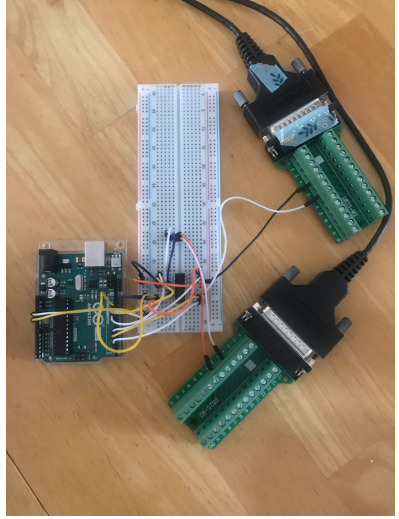

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Place the slide holder into the inner stage so that the mounting and pushing blocks align



Wiring the Arduino

<u>Step 1</u>	<u>Instructions</u>	<u>Pictures</u>
1	Break out DB25 cable and wire to 25 pin terminal block with aid of voltmeter	
2	Connect terminal block to actuator	
3	Connect bench top power supply to pin 5 (Motor +) and 7 (Motor -) using alligator clips	
4	Turn up the voltage until actuator rod moves in order to determine the minimum voltage required to operate the actuator	

5	Wire arduino, bread board, and the terminal block of each actuator according to Figure 1 below. Actuator #2 is wired as a mirror inverse of Actuator #1.	
6	Develop code in Arduino IDE and Upload to Arduino Uno via USB cable. Connect 10V power supply to arduino to test code.	

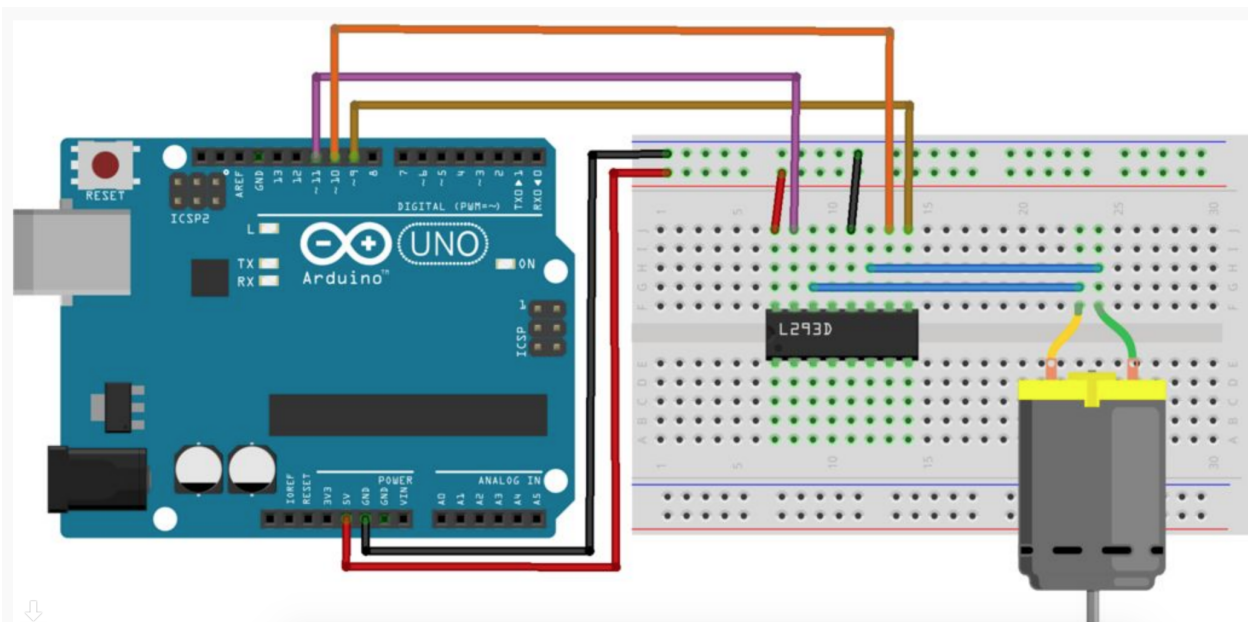


Figure 1. Arduino Uno Breadboard Wiring to control Motor #1. Add wires for Motor #2 in Mirror Inverse fashion.

Design History Record

MPI Steps	Part	Deviations from MPI	Completed By	Signature	Date
1-6	Outer Stage		Alex Schnorr	<i>Alex Schnorr</i>	3/1/19
1	Inner Stage		Theo Anastos	<i>Theo Anastos</i>	2/10/19
2-4	Inner Stage	In Step 3 the opening wasn't wide enough to fit the slide holder so a 1/2" endmill was used to increase the space in the part	Theo Anastos	<i>Theo Anastos</i>	2/22/19
5-7	Inner Stage		Theo Anastos	<i>Theo Anastos</i>	3/1/19
1-4	Slide Holder		Alex Schnorr	<i>Alex Schnorr</i>	2/22/19
5	Slide Holder		Theo Anastos	<i>Theo Anastos</i>	3/1/19
1-5	Hardware Assembly	The slide holder wasn't the right height to align the pushing block with the corresponding mounting block on the slide holder so metal spacers were used	Theo Anastos	<i>Theo Anastos</i>	3/1/19
1-4	Arduino		Ali Flesch	<i>Ali Flesch</i>	3/2/19

6.4 Divergence Between Final Design and Final Functional Prototype

Due to differences in height between the slide holder and inner stage, a metal spacer was necessary to ensure proper fitting of the mounting and pushing blocks. This metal spacer allowed the pushing block to fit concentric to the pushing rod of the linear actuator. Epoxy was used to fix the pushing and mounting blocks to each stage; tap drilling was not an option due to the risk of buckling of the thin metal. Although the slide holder was manufactured in the CNC, given dimensions did not fit the given microfluidic chip. An ¼” end mill was used to increase width and fillet radius by 0.05 in.

7.0 IQ/OQ

7.1 DOE

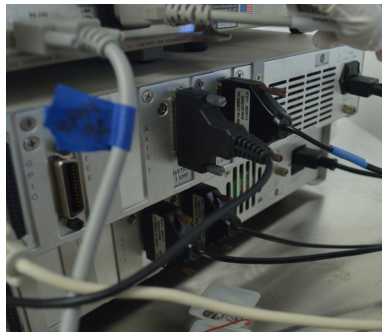
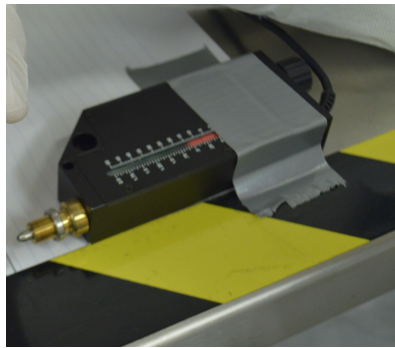
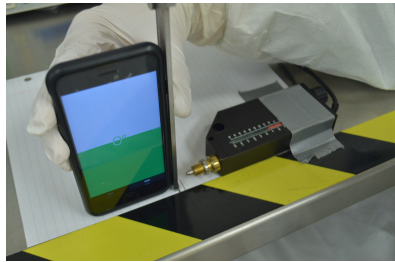
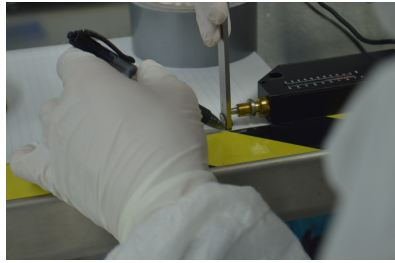
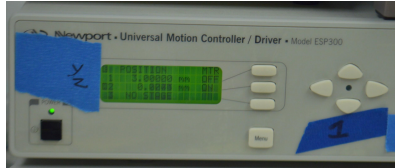
7.1.1 Displacement Capability

This pass or fail test is intended to confirm that the 850G Series Linear Actuators responsible for moving the microscope stage in the x and y direction are capable of extending at least 35 mm and 25 mm, respectively. This test was performed by plugging the actuator into the ESP300 Universal Motion Controller in the Microfabrication Laboratory. A sheet of paper was then lined against a straight edge and secured firmly with a piece of duct tape. After the paper was in place, the actuator was taped on top of the paper against the same straight edge. A ruler was then vertically lined against the actuator rod at its initial position and held at a 90 degree angle using a level. A pen was used to draw a line against the edge of the ruler to mark the initial position of the actuator rod. After marking the initial position, the ESP300 Universal Motion Controller was used to move the actuator rod to 35mm or 25mm, depending on the actuator. The final position of the actuator rod was marked using the procedure described above. Calipers were then used to measure the distance (d) between the initial and final position. If d was less than 35 mm or 25 mm, depending on the actuator, the test failed. This test was performed 10 times per actuator. The details of this experimental design and test plan are summarized below in Table VIII and IX, respectively.

Table VIII. Displacement Capability Experimental Design (Actuator #1 Newport / Actuator #2 Newport / Actuator #1 Arduino / Actuator #2 Arduino)

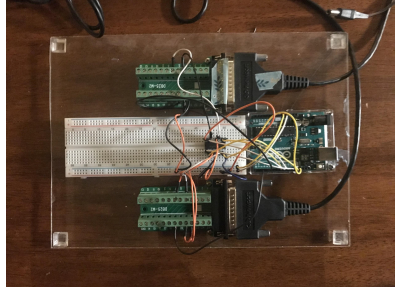
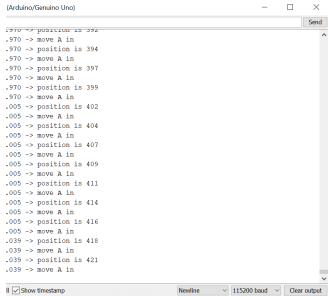
Engineering Metric	Specification	Test Location	Apparatus Training	Sample Size	Power
Displacement (d)	$d \geq 25 \text{ mm}$ in x-direction $d \geq 15 \text{ mm}$ in y-direction	Microfabrication Laboratory	Clean Room Protocol Experience with ESP300 Universal Motion Controller, Calipers, and JMP	N = 10 per direction	N/A

Table IX. Displacement Capability Newport Test Plan (Actuator #1 / Actuator #2)

Step	Description	Visual Depiction
1	Plug 850G Series Linear Actuator into ESP300 Universal Motion Controller in Microfabrication Laboratory.	
2	Line sheet of paper and actuator against straight edge and secure with duct tape.	
3	Line ruler against actuator rod at initial position. Ensure that ruler is at a 90 degree angle using a level.	
4	Use pen to draw a line against the edge of the ruler to mark initial position.	
5	Use ESP300 Universal Motion Controller to move actuator to 25 mm (X-direction actuator) or 15 mm (Y-direction actuator).	

6	Repeat Step 4 to mark final position.	
7	Use Calipers to measure the distance (d) between the initial and final position and record into Excel.	
9	Repeat Step 1-7 10 times per actuator.	

Table X. Displacement Capability Arduino Test Plan (Actuator #1 / Actuator #2)

Step	Description	Visual Depiction
1	Connect Actuator #1 and Actuator #2 to DB25 terminal blocks and 10V power supply to Arduino Uno	
2	Run code to move actuator #1 10,000 encoder turns (25mm)	
3	Watch serial monitor for actual encoder position at the end of the run	
4	Run code to move actuator #2 6,000 encoder turns (15mm)	
5	Watch serial monitor for actual encode position at the end of the run	
6	Ensure that all encoder positions are $\geq 10,000$ for Actuator #1 and $\geq 6,000$ for Actuator #2	

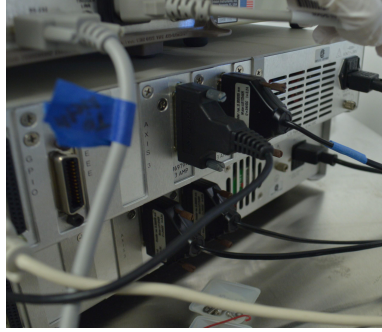
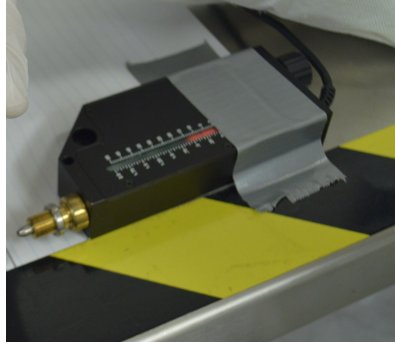
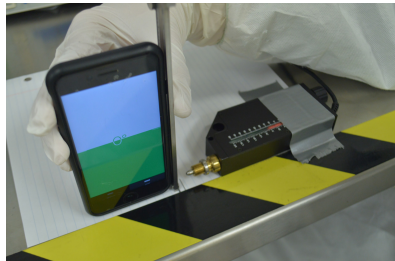
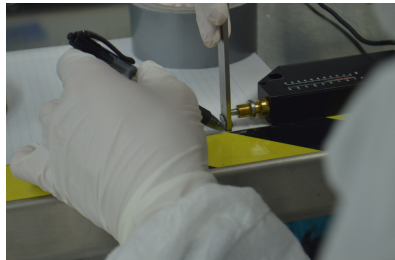

7.1.2 Position Accuracy

This test is intended to confirm that the 850G Series Linear Actuators responsible for moving the microscope stage in the x and y direction have a position accuracy within 5 microns. This test was performed by plugging the actuator into the ESP300 Universal Motion Controller in the Microfabrication Laboratory. A sheet of paper was then lined against a straight edge and secured firmly with a piece of duct tape. After the paper was in place, the actuator was taped on top of the paper against the same straight edge. A ruler was then vertically lined against the actuator rod at its initial position and held at a 90 degree angle using a level. A pen was used to draw a line against the edge of the ruler to mark the initial position of the actuator rod. After marking the initial position, the ESP300 Universal Motion Controller was used to move the actuator rod to 5mm. The final position of the rod was marked using the procedure described above. Calipers were then used to measure the distance (d) between the initial and final position. The difference (x) between d and the target position of 5mm was then calculated in JMP. This process was repeated 100 times. After collecting all of the data, a one-sample, upper-level t-test was conducted in JMP to determine whether x was significantly greater than the desired position accuracy of 5 mm. The details of this experimental design and test plan are summarized below in Table XI and XII, respectively.

Table XI. Position Accuracy Experimental Design (Actuator #1 Newport / Actuator #2 Newport / Actuator #1 Arduino / Actuator #2 Arduino)

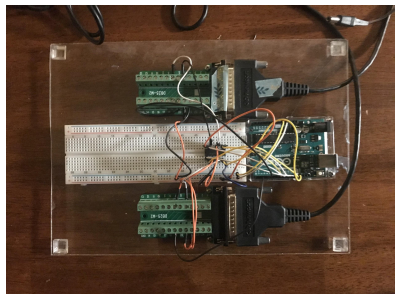
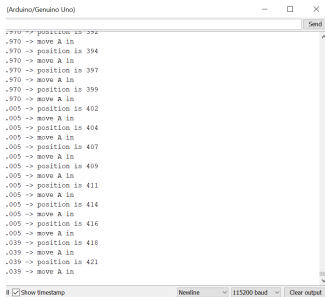
Engineering Metric	Specification	Test Location	Apparatus Experience/Training	Sample Size	Power
Accuracy	Actuator Position Accuracy within 5 microns: 1 sample, upper level t-test $p > 0.05$	Microfabrication Laboratory	Clean Room Protocol Experience with ESP300 Universal Motion Controller, Calipers, and JMP	N = 100	Actuator #1 Newport: 0.98 Actuator #2 Newport: 0.91 Actuator #1 Arduino: 0.92 Actuator #2 Arduino: 0.81

Table XII. Position Accuracy Newport Test Plan (Actuator #1 / Actuator #2)

Step	Description	Visual Depiction
1	Plug 850G Series Linear Actuator into ESP300 Universal Motion Controller in Microfabrication Laboratory.	
2	Line sheet of paper and actuator against straight edge and secure with duct tape.	
3	Line ruler against actuator rod at initial position. Ensure that ruler is at a 90 degree angle using a level.	
4	Use pen to draw a line against the edge of the ruler to mark initial position.	
5	Use ESP300 Universal Motion Controller to move actuator to 5 mm.	
6	Repeat Step 4 to mark final position.	

7	Use Calipers to measure the distance (d) between the initial and final position and record into JMP.	
9	Calculate the difference (x) between d and the target position of 5mm in JMP.	
10	Repeat step 1-9 100 times per actuator.	
11	Perform a one-sample, upper-level t-test in JMP to determine whether x was significantly greater than the desired position accuracy of 5 mm.	

Table XIII. Position Accuracy Arduino Test Plan (Actuator #1 / Actuator #2)

Step	Description	Visual Depiction
1	Connect Actuator #1 and Actuator #2 to DB25 terminal blocks and 10V power supply to Arduino Uno	
2	Run code to move actuator 2,000 encoder turns (5mm), 6,000 encoder turns (15mm), and 12,000 (30mm)	
3	Watch serial monitor for actual encoder position at the end of the run	
4	Record value into Excel (N = 30) and calculate difference between expected and actual final position (x)	
5	Copy x values into JMP. Perform a one sample, one-sided t-test comparing x to 5 microns	

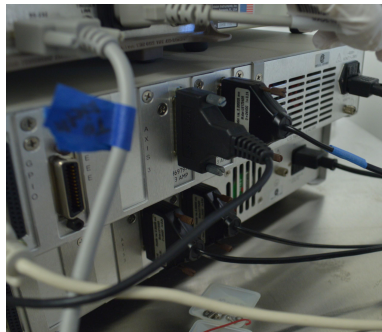
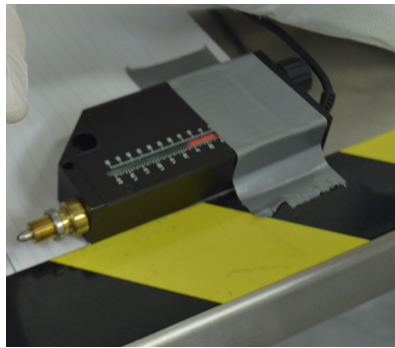
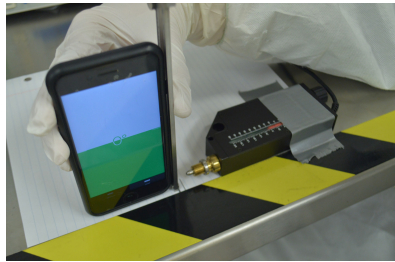
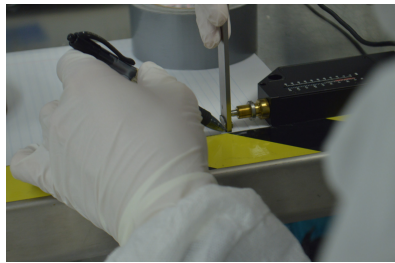
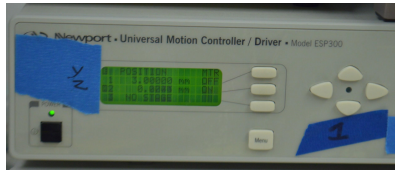
7.1.3 Position Error over Distance

This test is intended to confirm that the position accuracy of the 850G Series Linear Actuators responsible for moving the microscope stage in the x and y direction does not significantly fluctuate with distance traveled. This test was performed by plugging the actuator into the ESP300 Universal Motion Controller in the Microfabrication Laboratory. A sheet of paper was then lined against a straight edge and secured firmly with a piece of duct tape. After the paper was in place, the actuator was taped on top of the paper against the same straight edge. A ruler was then vertically lined against the actuator rod at its initial position and held at a 90 degree angle using a level. A pen was used to draw a line against the edge of the ruler to mark the initial position of the actuator rod. After marking the initial position, the ESP300 Universal Motion Controller was used to move the actuator rod to 5 mm (N=10), 15 mm (N=10), and 30 mm (N=10). The final position of the rod was marked using the procedure described above. Calipers were then used to measure the distance (d) between the initial and final position. The difference (x) between d and the target position was then calculated in JMP. After collecting all of the data, a one-way ANOVA was conducted in JMP to determine whether travel distance has a significant effect on x, or position error. The details of this experimental design and test plan are summarized below in Table XIV and XV, respectively.

Table XIV. Position Error over Distance Traveled Experimental Design (Actuator #1 Newport / Actuator #2 Newport / Actuator #1 Arduino / Actuator #2 Arduino)

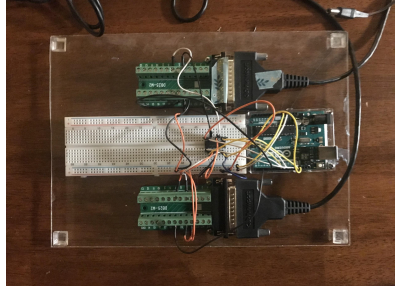
Engineering Metric	Specification	Test Location	Apparatus Experience/Training	Sample Size	Power
Accuracy	Actuator Position Accuracy does not significantly change with distance travelled: One-way ANOVA $p > 0.05$	Microfabrication Laboratory	Clean Room Protocol Experience with ESP300 Universal Motion Controller, Calipers, and JMP	N = 10 per travel distance	Actuator #1 Newport: 0.99 Actuator #2 Newport: 0.99 Actuator #1 Arduino: 0.97 Actuator #2 Arduino: 0.96

Table XV. Position Error over Distance Traveled Newport Test Plan (Actuator #1 / Actuator #2)

Step	Description	Visual Depiction
1	Plug 850G Series Linear Actuator into ESP300 Universal Motion Controller in Microfabrication Laboratory.	
2	Line sheet of paper and actuator against straight edge and secure with duct tape.	
3	Line ruler against actuator rod at initial position. Ensure that ruler is at a 90 degree angle using a level.	
4	Use pen to draw a line against the edge of the ruler to mark initial position.	
5	Use ESP300 Universal Motion Controller to move actuator to 5 mm (N = 10), 15 mm (N = 10), and 30 mm (N = 10).	

6	Repeat Step 4 to mark final position.	
7	Use Calipers to measure the distance (d) between the initial and final position and record into JMP.	
9	Calculate the difference (x) between d and the target position in JMP.	
10	Perform a one-way ANOVA in JMP to determine whether travel distance has a significant effect on x.	

Table XVI. Position Error over Distance Traveled Arduino Test Plan (Actuator #1 / Actuator #2)

Step	Description	Visual Depiction
1	Connect Actuator #1 and Actuator #2 to DB25 terminal blocks and 10V power supply to Arduino Uno.	
2	Run code to move actuator 2,000 encoder turns (5mm), 6,000 encoder turns (15mm), and 12,000 (30mm).	
3	Watch serial monitor for actual encoder position at the end of the run.	

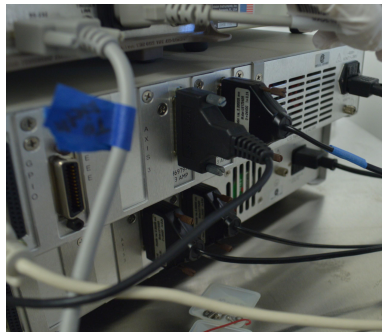
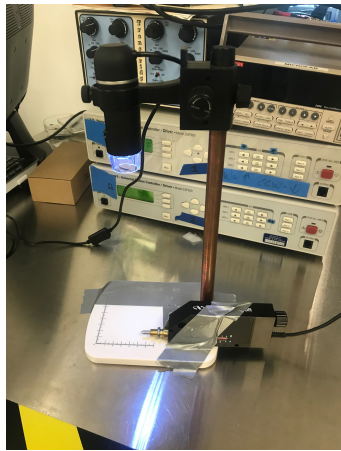
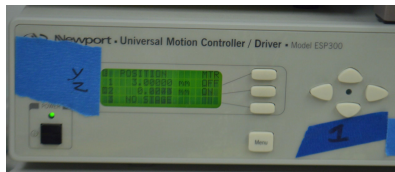
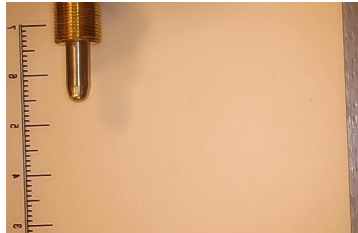
7.1.4 Position Return Repeatability

This test is intended to confirm that the position return repeatability of the 850G Series Linear Actuators responsible for moving the microscope stage in the x and y direction is within 1 micron. This test was performed by plugging the actuator into the ESP300 Universal Motion Controller in the Microfabrication Laboratory. The actuator was then securely taped on the surface of a Crenova Digital Microscope. The ESP300 Universal Motion Controller was used to move the actuator rod to 5 mm (Run #1), 10 mm, back to 5 mm (Run # 2), and then back to the origin. Using the Crenova Digital Microscope, a picture was taken of the actuator rod at each position. This process was repeated 100 times. All of the pictures taken at the end of Run #1 and Run #2 were then uploaded into ImageJ. Within ImageJ, the line tool was selected and positioned at (128,0). While holding down the shift key, the line was extended from (128, 0) to the end of the actuator rod. The command key and “M” were then held down simultaneously to add the measured length to the data pad. The measurements for Run #1 and Run #2 were then exported into JMP. A new formula column was created to calculate the difference in length between runs. A one sample, upper-level t-test ($N = 50$) was then performed to determine whether position repeatability was within 1 micron. The details of this experimental design and test plan are summarized below in Table XVII and XVIII, respectively.

Table XVII. Position Repeatability Experimental Design (Actuator #1 Newport / Actuator #2 Newport / Actuator #1 Arduino / Actuator #2 Arduino)

Engineering Metric	Specification	Test Location	Apparatus Experience/Training	Sample Size	Power
Repeatability	Position return repeatability within 1 micron: 1 sample, upper level t-test ($p > t$) > 0.05	Microfabrication Laboratory	Clean Room Protocol Experience with ESP300 Universal Motion Controller, Crenova Digital Microscope, ImageJ and JMP	$N = 50$	Actuator #1 Newport Newport: 0.99 Actuator #2 Newport: 0.99 Actuator #1 Arduino: 0.99 Actuator #2 Arduino: 0.99

Table XVIII. Position Return Repeatability Newport Test Plan (Actuator #1 / Actuator #2)

Step	Description	Visual Depiction
1	Plug 850G Series Linear Actuator into ESP300 Universal Motion Controller in Microfabrication Laboratory.	
2	Line actuator against straight edge of Crenova Digital Microscope	
3	Use ESP300 Universal Motion Controller to move actuator to 5 mm (Run #1), 10 mm, 5 mm (Run # 2), and 0 mm	
4	Use Crenova Digital Microscope to take a picture of the actuator at each position	

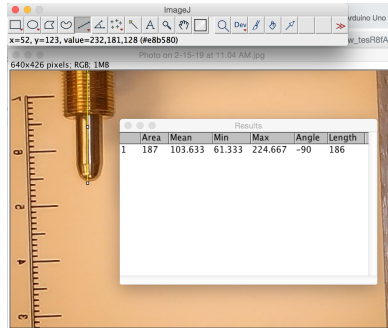
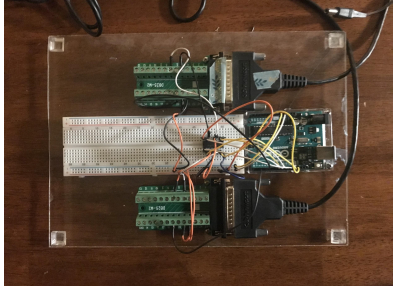
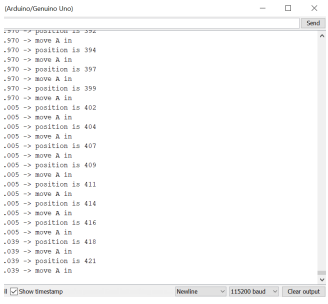
5	<p>Upload the images from Run #1 and Run #2 into ImageJ. Select the line tool and position it at (128,0). Hold the shift key and extend line to end of actuator rod. Press command + “M” to add length to data pad.</p>																																								
6	<p>Export data into JMP. Calculate the difference in length between Run #1 and Run #2. Perform a one sample, upper-level t-test (N = 50) to determine whether position repeatability was within 1 micron.</p>	<table><tr><th>Run #1</th><th>Run #2</th><th>Difference</th></tr><tr><td>188</td><td>187</td><td>1</td></tr><tr><td>188</td><td>188</td><td>0</td></tr><tr><td>187</td><td>187</td><td>0</td></tr><tr><td>187</td><td>187</td><td>0</td></tr><tr><td>187</td><td>187</td><td>0</td></tr><tr><td>187</td><td>187</td><td>0</td></tr><tr><td>187</td><td>187</td><td>0</td></tr><tr><td>187</td><td>187</td><td>0</td></tr><tr><td>186</td><td>187</td><td>1</td></tr><tr><td>187</td><td>186</td><td>1</td></tr><tr><td>186</td><td>186</td><td>0</td></tr><tr><td>186</td><td>186</td><td>0</td></tr></table>	Run #1	Run #2	Difference	188	187	1	188	188	0	187	187	0	187	187	0	187	187	0	187	187	0	187	187	0	187	187	0	186	187	1	187	186	1	186	186	0	186	186	0
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Table XIX. Position Return Repeatability Arduino Test Plan (Actuator #1 / Actuator #2)

Step	Description	Visual Depiction																																										
1	Connect Actuator #1 and Actuator #2 to DB25 terminal blocks and 10V power supply to arduino																																											
2	Run code to move actuator 2,000 encoder turns (Run #1) 4,000 encoder turns, 2,000 encoder turns (Run #2), and back to the origin.																																											
3	Watch serial monitor for actual encoder position at the end of Run #1 and Run #2 (N = 50).																																											
4	Import encoder positions into JMP and calculate the difference in length between Run #1 and Run #2 (x).																																											
5	Perform a one sample, upper level t-test to determine whether x was within 1 micron	<table border="1"> <thead> <tr> <th>Run #1</th><th>Run #2</th><th>Difference</th></tr> </thead> <tbody> <tr><td>188</td><td>187</td><td>1</td></tr> <tr><td>188</td><td>188</td><td>0</td></tr> <tr><td>187</td><td>187</td><td>0</td></tr> <tr><td>187</td><td>187</td><td>0</td></tr> <tr><td>187</td><td>187</td><td>0</td></tr> <tr><td>187</td><td>187</td><td>0</td></tr> <tr><td>187</td><td>187</td><td>0</td></tr> <tr><td>187</td><td>187</td><td>0</td></tr> <tr><td>187</td><td>187</td><td>0</td></tr> <tr><td>186</td><td>187</td><td>1</td></tr> <tr><td>187</td><td>186</td><td>1</td></tr> <tr><td>186</td><td>186</td><td>0</td></tr> <tr><td>186</td><td>186</td><td>0</td></tr> </tbody> </table>	Run #1	Run #2	Difference	188	187	1	188	188	0	187	187	0	187	187	0	187	187	0	187	187	0	187	187	0	187	187	0	187	187	0	186	187	1	187	186	1	186	186	0	186	186	0
Run #1	Run #2	Difference																																										
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7.2 Verification and Validation

7.2.1 Actuator #1 Displacement Capability Test Results

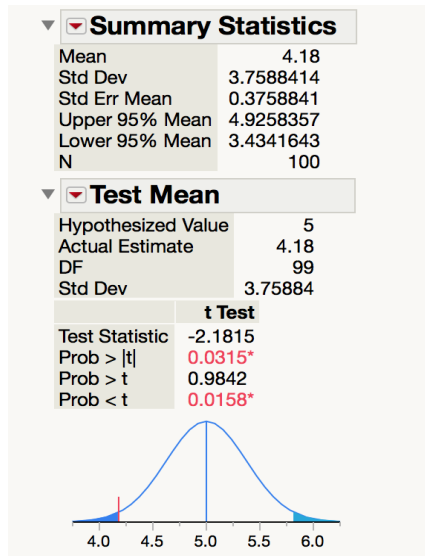
As shown below in Table XX, Actuator #1 passed the displacement capability test for all ten runs.

Table XX. Actuator #1 Displacement Capability Test Results

Displacement ▼	Rest ▼
> 35mm	PASS
> 35mm	PASS
> 35mm	PASS
> 35mm	PASS
> 35mm	PASS
> 35mm	PASS
> 35mm	PASS
> 35mm	PASS
> 35mm	PASS
> 35mm	PASS

7.2.2 Actuator #1 Position Accuracy Test Results

The JMP output for the one sample, upper-level Actuator #1 Position Accuracy t-test is displayed below. Due to a large ($p > t$) value ($0.9842 > 0.05$), there is insufficient evidence to suggest that position accuracy is not within 5 microns. Therefore, per the experimental design specifications outlined in Table XI, the position accuracy of Actuator #1 aligns with the customer requirements.



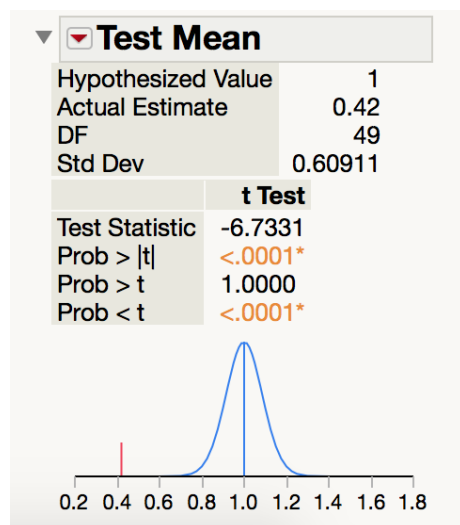
7.2.3 Actuator #1 Position Error over Distance Traveled Test Results

The JMP output for the one-way ANOVA Actuator #1 Position Error Over Distance Traveled test is displayed below. Due to a large p-value ($0.2713 > 0.05$), there is insufficient evidence to suggest that position error significantly fluctuates with actuator travel distance. Therefore, per the experimental design specifications outlined in Table XIV, the position error of Actuator #1 as a function of displacement aligns with the customer requirements.

▼ Analysis of Variance				
Source	DF	Sum of Squares	Mean Square	F Ratio
Model	2	3.266667	1.63333	1.3696
Error	27	32.200000	1.19259	Prob > F
C. Total	29	35.466667		0.2713

7.2.4 Actuator #1 Position Return Repeatability Test Results

The JMP output for the one sample, upper-level Actuator #1 Position Return Repeatability t-test is displayed below. Due to a large ($p > t$) value ($1.00 > 0.05$), there is insufficient evidence to conclude that position return repeatability is not within 1 micron. Therefore, per the experimental design specifications outlined in Table XVII, the position return repeatability of Actuator #1 satisfies the customer requirements.



7.2.5 Actuator #2 Displacement Capability Test Results

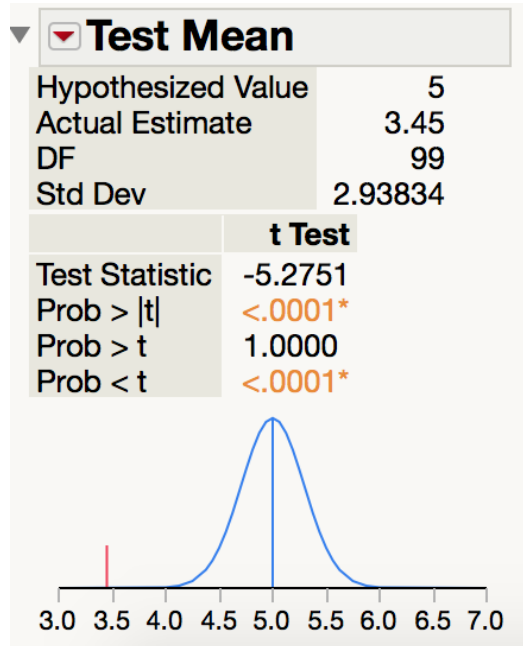
As shown below in Table XXI, Actuator #2 passed the displacement capability test for all ten runs.

Table XXI. Actuator #1 Displacement Capability Test Results

Displacement ▼	Rest ▼
> 25mm	PASS
> 25mm	PASS
> 25mm	PASS
> 25mm	PASS
> 25mm	PASS
> 25mm	PASS
> 25mm	PASS
> 25mm	PASS
> 25mm	PASS
> 25mm	PASS

7.2.6 Actuator #2 Position Accuracy Test Results

The JMP output for the one sample, upper-level Actuator #2 Position Accuracy t-test is displayed below. Due to a large ($p > t$) value ($1.0000 > 0.05$), there is insufficient evidence to suggest that position accuracy is not within 5 microns. Therefore, per the experimental design specifications outlined in Table XI, the position accuracy of Actuator #2 aligns with the customer requirements.



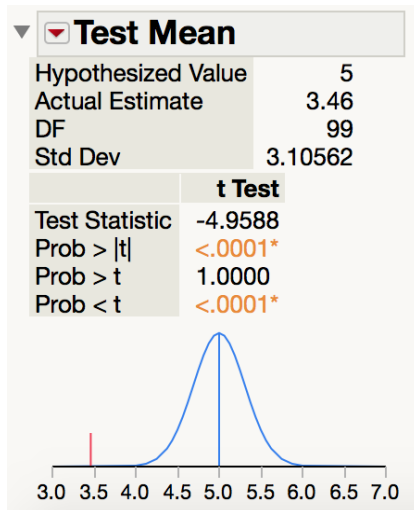
7.2.7 Actuator #2 Position Error over Distance Traveled Test Results

The JMP output for the one-way ANOVA Actuator #2 Position Error Over Distance Traveled test is displayed below. Due to a large p-value ($0.6528 > 0.05$), there is insufficient evidence to suggest that position error significantly fluctuates with actuator travel distance. Therefore, per the experimental design specifications outlined in Table XIV, the position error of Actuator #2 as a function of displacement aligns with the customer requirements.

Analysis of Variance					
Source	DF	Sum of Squares	Mean Square	F Ratio	Prob > F
Distance Traveled (mm)	2	0.866667	0.43333	0.4333	0.6528
Error	27	27.000000	1.00000		
C. Total	29	27.866667			

7.2.10 Actuator #1 Arduino Position Accuracy Test Results

The JMP output for the one sample, upper-level Arduino Actuator #1 Position Accuracy t-test is displayed below. Due to a large ($p > t$) value ($1.0000 > 0.05$), there is insufficient evidence to suggest that the position accuracy of Actuator #1 is not within 5 microns. Therefore, per the experimental design specifications outlined in Table XI, the position accuracy of Actuator #1 aligns with the customer requirements.



7.2.11 Actuator #1 Arduino Position Error over Distance Traveled Test Results

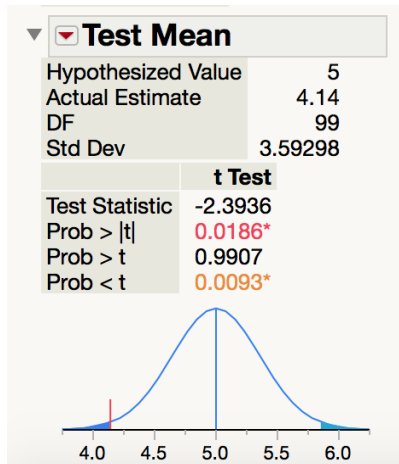
The JMP output for the one-way ANOVA Fully Assembly Actuator #1 Position Error Over Distance Traveled test is displayed below. Due to a large p-value ($0.8499 > 0.05$), there is insufficient evidence to suggest that inner stage position error significantly fluctuates with actuator travel distance. Therefore, per the experimental design specifications outlined in Table XIV, the position error of Actuator #1 as a function of displacement aligns with the customer requirements.

Analysis of Variance

Source	DF	Sum of Squares	Mean Square	F Ratio
Model	2	0.466667	0.23333	0.1636
Error	27	38.500000	1.42593	Prob > F
C. Total	29	38.966667		0.8499

7.2.14 Actuator #2 Arduino Position Accuracy Test Results

The JMP output for the one sample, upper-level Position Accuracy t-test is displayed below. Due to a large ($p > t$) value ($1.0000 > 0.05$), there is insufficient evidence to suggest that the position accuracy of the slide holder is not within 5 microns. Therefore, per the experimental design specifications outlined in Table XI, the position accuracy of Actuator #2 aligns with the customer requirements.



7.2.15 Actuator #2 Arduino Position Error over Distance Traveled Test Results

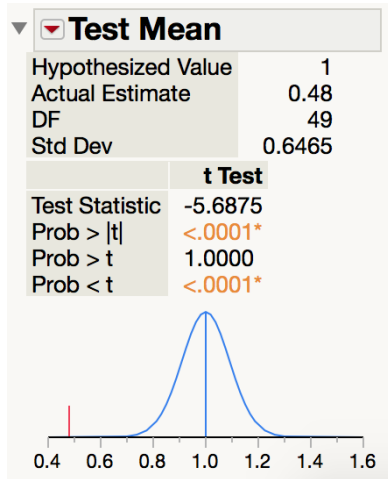
The JMP output for the one-way ANOVA Position Error Over Distance Traveled test is displayed below. Due to a large p-value ($0.6161 > 0.05$), there is insufficient evidence to suggest that slide holder position error significantly fluctuates with actuator travel distance. Therefore, per the experimental design specifications outlined in Table XIV, the position error of Actuator #2 as a function of displacement aligns with the customer requirements.

Analysis of Variance

Source	DF	Sum of Squares	Mean Square	F Ratio
Model	2	0.800000	0.400000	0.4932
Error	27	21.900000	0.811111	Prob > F
C. Total	29	22.700000		0.6161

7.2.16 Actuator #2 Arduino Return Repeatability Test Results

The JMP output for the one sample, upper-level Arduino Actuator #2 Position Return Repeatability t-test is displayed below. Due to a large ($p > t$) value ($1.00 > 0.05$), there is insufficient evidence to conclude that position return repeatability is not within 1 micron. Therefore, per the experimental design specifications outlined in Table XVII, the position return repeatability of Actuator #2 satisfies the customer requirements.



8.0 Conclusions and Recommendations

8.1 Recommendations

Full functionality of the system will be completed when the camera system is fully synchronized with the system. This step of the process was unable to be completed during the time frame of the project due to lack of access and training of the SVM-340. Although this prototype was successful in short-term testing, further testing should be completed to ensure that the CellOptimizer is able to support microfluidic cell systems over time. Collection of long-term run data will reveal steps that must be taken to ensure proper system maintenance, particularly with effects of cyclic loading on function of the rubber band; steps can then be taken to prevent malfunction of the system. Because the system is made of aluminum, thermal analysis should be completed to ensure that the heating system is minimally affected by stage. Finally, a long-term biocompatibility test should be completed to ensure the final goal of long-term viability of cells is achieved.

8.2 Conclusion

The *CellOptimizer* automated microscope stage was successful in meeting the objectives outlined in the Indications for Use, design specification matrix, and design of experiments. The system was built from the ground up: designed in SolidWorks, rapid prototyped, manufactured from aluminum, automated with an Arduino microcontroller, and validated through experimental testing. The automated stage passed all tests, concluding that customer requirement-based specifications were met. The system met specifications of a displacement greater than 25 mm and 35 mm in respective translational directions. Statistically significant repeatability return and accuracy functions indicate that the system satisfies the customer's need for long-term imaging of microfluidic chip wells. The system also satisfies the requirement for Newport 850G Linear Actuators to interface with an Arduino controller instead of the given ESP3000 Controller. Although the CellOptimizer may require additional testing to achieve specific end goals beyond the scope of this capstone project, modifications to the device should be relatively simple.

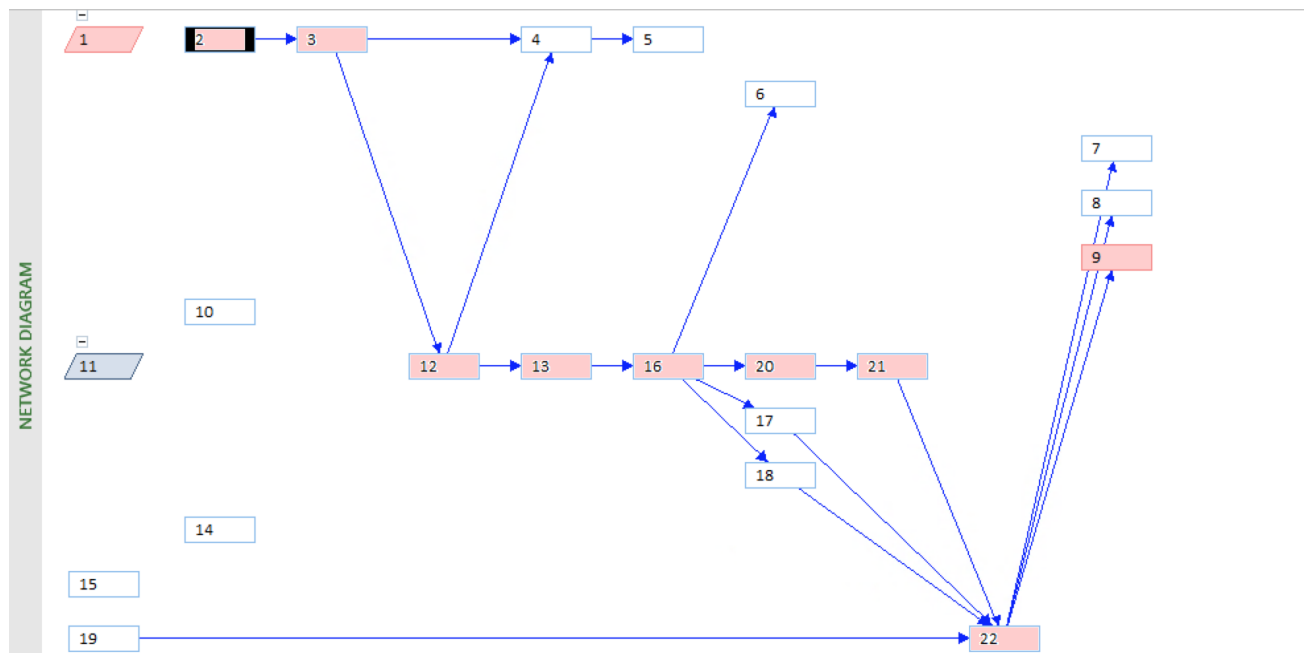
9.0 Acknowledgments

We would like to acknowledge and thank Dr. Hawkins, Dr. Heylman, and Dr. Whitt for providing resources, advice, and training throughout the duration of the project. We would also like to acknowledge the benefactor of the Hannah Forbes Fund for providing us with the resources necessary to complete this device.

10.0 Appendices10.1 Appendix A: References10.2 Appendix B: Project Plan (PERT Chart)

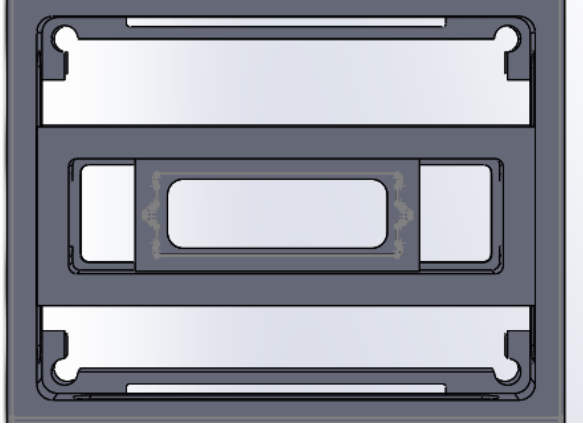
Task Number	Task Name	Duration	Start	End	Predecessors
1	Presentations	126 days	Mon 10/1/18	Wed 3/20/19	
2	Project Planning Presentation	13 days	Mon 10/1/18	Wed 10/17/18	
3	Concept Review presentation	26 days	Thu 10/18/18	Thu 11/22/18	2
4	Preliminary Design/Design Freeze Presentation	21 days	Thu 12/13/18	Thu 1/10/19	3,12
5	Final Test & Manufacturing Plan Presentation	16 days	Fri 1/11/19	Fri 2/1/19	4
6	Preliminary Functional Prototype Presentation	11 days	Wed 1/30/19	Wed 2/13/19	16
7	Final Poster Presentation	1 day	Mon 3/11/19	Mon 3/11/19	22
8	Final Design & Prototype Presentation	1 day	Mon 3/11/19	Mon 3/11/19	22
9	Final Report	8 days	Mon 3/11/19	Wed 3/20/19	22
10	Case Study & Debrief	1 day	Mon 3/18/19	Mon 3/18/19	
11	Phase I (Fall)	60 days?	Mon 10/1/18	Fri 12/21/18	
12	CAD Designs	14 days	Fri 11/23/18	Wed 12/12/18	3
13	Rapid Prototype	7 days	Thu 12/13/18	Fri 12/21/18	12
14	Yellow Tag	35 days?	Mon 10/1/18	Fri 11/16/18	
15	Phase II (Winter)	56 days	Mon 10/1/18	Mon 12/17/18	
16	3D Prototyping	14 days	Thu 1/10/19	Tue 1/29/19	13
17	CNC	1 day	Fri 2/22/19	Fri 2/22/19	16

18	Mill Inner Stage	10 days	Sat 2/16/19	Thu 2/28/19	16
19	Accuracy/Repeatability Test (Actuator)	1 day	Wed 2/13/19	Wed 2/13/19	
20	Actuator Electrical Testing	8 days	Mon 2/18/19	Wed 2/27/19	16
21	Electrical Setup	1 day	Sat 3/2/19	Sat 3/2/19	20
22	Accuracy/Repeatability Test (Assembly)	1 day	Sun 3/3/19	Sun 3/3/19	21,19,17,18



10.3 Appendix C: CAD Drawings

10.3.1 Fully Assembled CAD Design



10.4 Appendix D: FMEA, Hazard & Risk Assessment

10.4.1 FMEA

Overall, there are many components whose failure may have a significant effect on the microscopy system. Even failures in small parts could have an effect on the alignment of the microfluidic chip, which would affect the outcome of the user's experiment. Since this automated process does not require user presence, it is especially important to predict wearing of parts or equipment failure. Proper training should be given to users in the areas of microscope use, device use, and microfluidic chip handling to decrease risk of component damage. A customer service contact should be available to quickly remedy any software or microscope hardware problems.

Component Name	Possible Failure Mode	Type	Cause of Failure	OC C	DET	SEV	RPN	Effect of Failure on System	Failure Improvement Alternative Actions (actions to fix the problem...)
Arduino	Electrical Short	M	Fluid Damage	1	1	10	10	No images would be captured	Replace or Protective Cover
Servo Motor	Worn	W	Overuse	2	1	10	20	No images would be captured	Replace
SVM340	Damaged	M	Power Outage	3	1	10	30	No images would be captured	Customer Support
Microfluidic Chip	Damaged	C	Customer Abuse	1	1	10	10	Samples ruined	Acquire new samples
VGA Resolution Analog CCD camera	Damaged	M	Customer Abuse	1	1	10	10	No images would be captured	Customer Support
Illuminator Module	Worn	W	Overuse	1	1	5	5	Image clarity decreased	Replace LEDs
Microscope Stage	Damaged	M	Customer Abuse	1	1	6	6	Samples not aligned properly	Customer Support
Stainless Steel Plate	Damaged	M	Customer Abuse	1	8	1	8	Samples not aligned properly	Customer Support
Fluorescent Light	Bulbs wear out	W	Overuse	1	1	3	3	No fluorescence	Replace bulbs
Base Stand	Damaged	C	Customer Abuse	2	1	5	10	Samples not aligned properly	Customer Support
Uscope Software	Error	M	Software Error	3	1	10	30	Microscope camera shuts down	Reinstall Software
Objectives	Damaged	M	Customer Abuse	1	1	7	7	No images would be captured	Clean/replace Objectives
9-pin D Sub Connector	Damaged	M	Physical Damage	1	1	5	5	No way to edit code	Replace

Microscope Lens	Damaged	C	Customer Abuse	1	1	7	7	No images would be captured	Clean/replace Lens
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10.4.2 Safety Hazard Checklist

1. Will any part of the design create hazardous revolving, reciprocating, running, shearing, punching, pressing, squeezing, drawing, cutting, rolling, mixing or similar action, including pinch points and shear points? **Y**
2. Can any part of the design undergo high accelerations/decelerations? **N**
3. Will the system have any large moving masses or large forces? **N**
4. Will the system produce a projectile? **Y**
5. Would it be possible for the system to fall under gravity creating injury? **Y**
6. Will a user be exposed to overhanging weights as part of the design? **N**
7. Will the system have any sharp edges? **N**
8. Will any part of the electrical systems not be grounded? **N**
9. Will there be any large batteries or electrical voltage in the system above 40 V? **N**
10. Will there be any stored energy in the system such as batteries, flywheels, hanging weights or pressurized fluids? **N**
11. Will there be any explosive or flammable liquids, gases, or dust fuel as part of the system? **N**
12. Will the user of the design be required to exert any abnormal effort or physical posture during the use of the design? **N**
13. Will there be any materials known to be hazardous to humans involved in either the design or the manufacturing of the design? **Y**
14. Can the system generate high levels of noise? **N**
15. Will the device/system be exposed to extreme environmental conditions such as fog, humidity, cold, high temperatures, etc? **N**
16. Is it possible for the system to be used in an unsafe manner? **N**
17. Will there be any other potential hazards not listed above? If yes, please explain on reverse.
N

10.4.3 Risk Assessment

With the use of potentially toxic organic reagents used within the microfluidic chip, it is essential that all system users are properly trained and obey all lab safety standards when handling chips. Use of isopropyl alcohol will ensure that none of the biological samples are contaminated, which would affect experimental outcomes. The system itself will use a linear actuator to drive an inner stage, causing pinch and shear points. These points can be factored into the design to protect users. The system could potentially fall off the microscope and cause minor injury, so the stage will be secured to the microscope pegs.

Description of Hazard	Planned Corrective Action	Planned Date	Actual
Exposure to organic reagents in microfluidic chip	Use of gloves when handling chip, wipe with IPA before/after use, and proper training	3/10/19	3/10/19
Pinch/shear points from moving actuator	Design microscope stage to protect user from actuator pinch/shear points	W 19	2/22/19
Falling stage	Attach stage to microscope securely (Pegs)	W 19	2/22/19
Rubber band projectile	Replace rubber bands on a regular basis. Use strong epoxy to ensure pushing/mounting blocks do not dislodge	W 19	3/1/19

10.5 Appendix E: Pugh Chart

Looking at the results from the first Pugh Chart, it was concluded that both Concept 2 and Concept 3 were superior to Concept 1. Then the second and third Pugh Charts showed that Concept 2 was superior to Concept 3. We ended up deducing that Concept 2 was the superior design. Although it is the most expensive and time consuming to produce, the advantages over the other two designs make it worth the cost and manufacturing time. Concept 2 provides greater adjustability, actuator surface area contact, stability, usability, and aesthetic. Concept 3, although lightweight and easy to produce, is the least aesthetic and stable design. Concept 3 also does not offer the large actuator surface area contact that Concept 2 provides. Taking into consideration these Pugh Charts as well as Dr. Hawkins customer specifications, we initially moved forward with Concept 2.

10.5.1 Pugh Chart Concept 1

Selection Criteria		Concepts		
		1	2	3
Weight		Datum	S	+
Cost			-	+
Adjustability			+	S
Volume			S	S
Actuator surface area contact			+	S
Manufacturing time			-	+
Stability			+	-
Aesthetic			S	-
Usability			+	S
Number of components			-	+
# of Pluses			4	4
# of Minuses			3	2

10.5.2 Pugh Concept #2

Selection Criteria		Concepts		
		2	1	3
Weight		Datum	S	+
Cost			+	+
Adjustability			-	-
Volume			S	S
Actuator surface area contact			-	-
Manufacturing time			+	+
Stability			-	-
Aesthetic			S	-
Usability			-	-
Number of components			+	+
# of Pluses			3	4
# of Minuses			4	5

10.5.3 Pugh Concept #3

Selection Criteria		Concepts		
		3	1	2
Weight		Datum	-	-
Cost			-	-
Adjustability			S	+
Volume			S	S
Actuator surface area contact			S	+
Manufacturing time			-	-
Stability			+	+
Aesthetic			+	+
Usability			S	+
Number of components			-	-
# of Pluses			2	5
# of Minuses			4	4

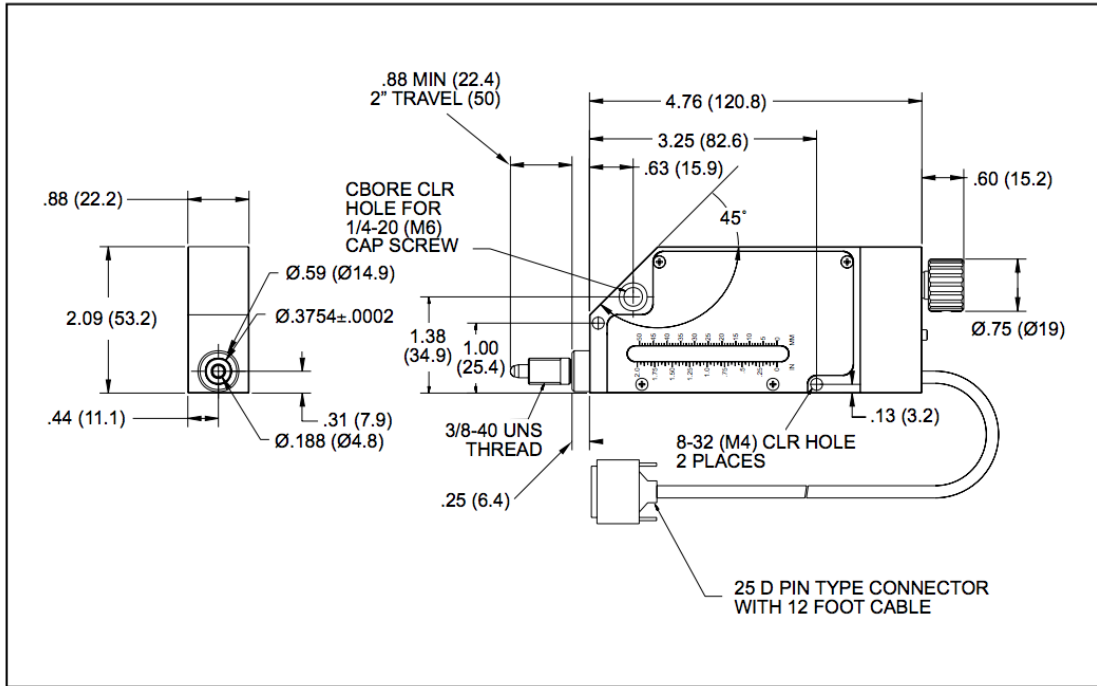
10.6 Appendix F: Vendor Information, Specifications, and Data Sheets

10.6.1 850G Series Linear Actuator

10.6.1.1 Specifications

Encoder Resolution		Part Number
Standard Actuators:	0.05101µm	850G, 850GV6
High Speed Actuators:	0.60514µm	850G-HS
Low Speed Actuators:	0.007985µm	850G-LS
Nominal Gearbox Ratio and Maximum Speed		
Standard Actuators:	262:1 ratio (1624 motor); 500µm/sec.	
High Speed Actuators:	22:1 ratio (1624 motor); 6000µm/sec.	
Low Speed Actuators:	1670:1 ratio (1516 motor); 78µm/sec.	
Backlash	< 20 micron typical with external load of 2 lbs.(1 kg) minimum	
Accuracy	< 0.1% of travel, cumulative	
Bi-directional	Repeatability: Better than 1 micron when backlash is compensated by controller (standard actuators) *1	
Encoder	Magnetic, 2KHz; open collector, quadrature output, +5V to +12V supply	
Absolute cyclic pitch	Error < 1 micron	
Time to reach full speed	< 50 msec at max. speed and acceleration settings	
Max. Side Load	5 lb. (2.3 kg) at full shaft extension	
Max. Axial Load	18 lb. (8 kg) standard and low speed actuators	
Cable	12 foot (3.6 m) cable integral to actuator terminated with 25-pin male Dsub connector	

10.6.1.2 Drawings



10.6.3. Vendor Information

Vendor	Newport Corporation
Headquarters	Irvine, CA
Founded	1969
Parent Organization	MKS Instruments, Inc.
Description	<p>“A leading global supplier of advanced technology products and systems to customers in scientific research, microelectronics...</p> <p>Newport has over 47 years of industry knowledge and expertise across a broad range of technologies” [1]</p>

10.6.2 ESP300 Motion Controller/Driver

10.6.2.1 Specifications

1.4.2 Specifications Function:

- Integrated motion controller and driver.

Number of motion axes:

- 1 to 3, in any combination or order of 2 phase stepper and brush DC motors, up to 48VDC, 3A per axis.

Trajectory type:

- Trapezoidal velocity profile
- S-curve velocity profile.

Motion device compatibility:

- Family of motorized Newport motion devices, using either stepper or DC motors
- Custom motion devices (call for compatibility).

DC motor control:

- 18 bit DAC resolution
- 4 MHz maximum encoder input frequency
- Digital PIDFF servo loop, 0.4 ms update rate.

Stepper motor control:

- Up to 1000 microstep resolutions per full step.

Computer Interface:

- RS232-C, 19200 baud, 8 bits, 8, N, 1
- IEEE-488 – optional, Please contact Newport for information

Utility interfaces:

- 16 bit digital inputs/outputs, user definable, in blocks of 8.
- Remote motor off input (interlock).

User Memory:

- 64 KB non-volatile program memory
- 512 byte command buffer

Operating modes:

- Local mode – stand-alone operation, executing motion from the front panel
- Remote mode – executing commands received over one of the computer interfaces or the optional handheld keypad
- Program execution mode – execution of a stored program.

Optional display:

- 80 character alpha-numeric LCD display
- Displays position, status, utility menus and setup screens.

Dimensions:

- 3" (2U) H x 16.5" W x 12" D (75 x 412 x 300 mm) for ESP300-XXXXX1 Models
- 3" (2U) H x 16.5" W x 13.5" D (75 x 412 x 342.9 mm) for ESP300-XXXXX2 Models

Power requirements:

- 100-240VAC $\pm 10\%$, 50/60 Hz
- 4A max for ESP300 - XXXXX1 Models
6.3A max for ESP300 - XXXXX2 Models

Fuses:

- T4A / 250VAC for XXXXX1 Models
- T6.3A/250VAC for XXXXX2 Models

Weight:

- 10.90Lbs. max (4.9kg) max for ESP300 - XXXXX1 Models
- 14.05Lbs. max (6.37kg) max for ESP300 - XXXXX2 Models

Operating conditions:

- Temperature: 0°C to 40°C
- Humidity: 20% to 90% RH, non-condensing

Other :

- Pollution degree :2
- Installation category: II
- Altitude: <2000m
- Instrument use: the model ESP300 is intended for indoor use only.

10.6.2.2 Vendor Information

See Section 10.6.1.3

10.6.3 6061-T651 Aluminum Plate

10.6.3.1 Specifications

6061 COMPOSITION	
ALUMINUM	& alloying elements:
SILICON	0.4-0.8%
MAGNESIUM	0.8-1.2%
IRON	0.7% (max)
ZINC	0.25% (max)
COPPER	0.15%-0.4%
MANGANESE	0.15% (max)
TITANIUM	0.15% (max)
CHROMIUM	0.04-0.35%
OTHER:	0.05% (max) per
	Other total: 0.15%

6061-T651 CHARACTERISTICS	
CORROSION RESISTANCE	B
STRESS-CORROSION CRACKING RESISTANCE	A
WORKABILITY	C
MACHINABILITY	C
BRAZEABILITY	A
WELDABILITY: GAS	A
WELDABILITY: ARC	A
WELDABILITY: RESISTANCE SPOT & SEAM	A

10.6.3.2 Vendor Information

Vendor	Midwest Steel and Aluminum
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10.6.4 Arduino Uno

10.6.4.1 Specifications

Microcontroller	ATmega328P
Operating Voltage	5V
Input Voltage (recommended)	7-12V
Input Voltage (limit)	6-20V
Digital I/O Pins	14 (of which 6 provide PWM output)
PWM Digital I/O Pins	6
Analog Input Pins	6
DC Current per I/O Pin	20 mA
DC Current for 3.3V Pin	50 mA
Flash Memory	32 KB (ATmega328P) of which 0.5 KB used by bootloader
SRAM	2 KB (ATmega328P)
EEPROM	1 KB (ATmega328P)
Clock Speed	16 MHz
LED_BUILTIN	13
Length	68.6 mm
Width	53.4 mm
Weight	25 g

10.6.4.2 Vendor Information

Vendor	Arduino
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10.6.5 L923D Motor Driver

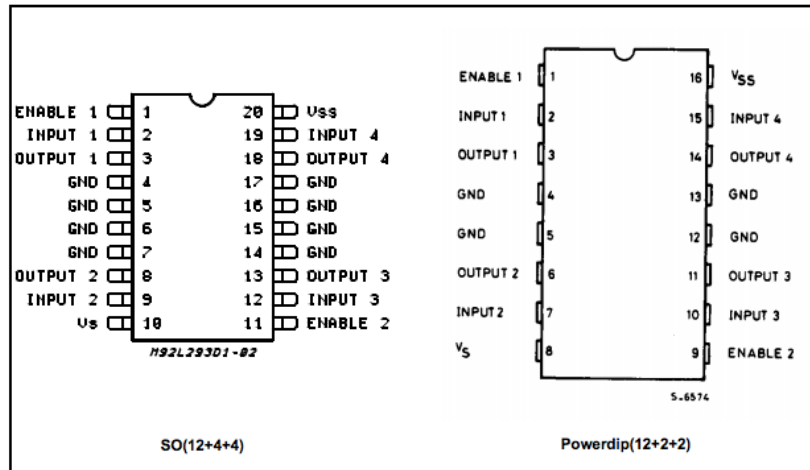
10.6.5.1 Specifications

L293D - L293DD

ABSOLUTE MAXIMUM RATINGS

Symbol	Parameter	Value	Unit
V_S	Supply Voltage	36	V
V_{SS}	Logic Supply Voltage	36	V
V_i	Input Voltage	7	V
V_{en}	Enable Voltage	7	V
I_o	Peak Output Current (100 μ s non repetitive)	1.2	A
P_{tot}	Total Power Dissipation at $T_{pins} = 90^\circ\text{C}$	4	W
T_{stg}, T_j	Storage and Junction Temperature	- 40 to 150	$^\circ\text{C}$

PIN CONNECTIONS (Top view)



THERMAL DATA

Symbol	Description	DIP	SO	Unit
$R_{th(j-pins)}$	Thermal Resistance Junction-pins	max.	14	$^\circ\text{C/W}$
$R_{th(j-amb)}$	Thermal Resistance junction-ambient	max.	80	$^\circ\text{C/W}$
$R_{th(j-case)}$	Thermal Resistance Junction-case	max.	14	$^\circ\text{C/W}$

(*) With 6sq. cm on board heatsink.

10.6.5.1 Vendor Information

Vendor	DigiKey Electronics
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10.7 Appendix G: Budget

Component	Budget
Hardware	\$506
Software	\$66
Testing	\$30
Total	\$602